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Potential of Pulse Flours as Partial Meat Replacers in Heat-Treated Emulsion-Type Meat Sausages

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Reformulation approaches in the meat industry are required to promote nutritional improvement, health functionality, and reduce environmental impact. A relevant approach among these is to reduce the amount of meat in meat products. Reduced-meat products should maintain or improve the sensory characteristics and nutritive value compared to conventional meat products. Among meat products, heat-treated emulsion-meat sausages are widely consumed and especially suitable for reformulation approaches. Due to its high protein content, with high functionally and biological value, pulse flour has a high potential to be used as meat replacer. Most studies regarding the replacement of meat with pulses have been made on fresh meat preparations where amounts of up to 15% of pulse flour did not negatively affect sensory quality while increased yield and firmness. However, studies using pulse flour in emulsion-type sausages are scarce. Further research is warranted to optimize the reformulation of these meat products using flour pulses. The topics to be addressed are the following: effects of pulse type, pulse pretreatments, such as soaking or germination, pulse flour treatments before incorporation into the meat mix, combination of pulses with other proper ingredients, and heat treatment intensity on the pulse antinutrient inactivation and the technological and edible quality traits of the pulse-containing sausages.

Keywords: dry-grain legumes, reformulation, extenders, Frankfurters, meat reduction

INTRODUCTION

A large part of the meat consumed in industrialized countries is previously processed. Versatility, more attractive products, improved cost-effectiveness, and higher shelf life are some of the main features that distinguish meat derivatives from fresh meat. There are many types meat derivatives, which are classified according to whether they are or are not heat treated or whether they are made with whole pieces of meat or minced meat.

Among the last, a relevant type of meat products is that composed of the heat-treated emulsion-type sausages, such as Frankfurters or hot dogs. Heat-treated emulsion-type sausages are made from cooking a homogeneous fine batter, i.e., finely cut meat, fatty tissue, and water, where the particles of meat or fat tissue are not visible, stuffed into casings (Feiner, 2006). The batter consists mainly of immobilized water, dissolved compounds such as common salt, phosphates, or curing agents, protein aggregates, suspended muscle, connective and fat tissue tiny particles, spices particles, and air micro bubbles. The particle size reduction in this type of meat product is carried out by cutting and shearing meat and non-meat ingredients using cutters or colloid mills at temperatures usually lower than 12°C.

Meat is considered a relevant dietary source of a wide range of nutrients, such as high-quality proteins, zinc, iron, selenium, and B12 vitamins (Salter, 2018; Cocking et al., 2020). On the other hand, an excess of meat and meat products in the diet is seem a problem in rich countries due to meat contributing to a diet with high fat, and cholesterol contents, unfavorable fatty acid profile, and the eventual presence of nitrites as additives (Grasso et al., 2014). Evidence from medical research indicated that long-term excessive meat intake, particularly red meat and processed meat, has been associated with increased risk of total mortality due to chronic diseases, such as cardiovascular disease, type 2 diabetes, and certain types of cancer such as colon cancer (De Smet and Vossen, 2016; Domingo and Nadal, 2017; Ekmekcioglu et al., 2018).

Concerns on excessive meat consumption are not only based on human health issues but also on the negative environmental impacts of intensive farming (Kowalski, 2019; Pintado and Delgado-Pando, 2020). Farmland used for livestock feed production, nitrogen emissions, and greenhouse gas emissions has been estimated to be reduced by 20, 40, and 25–40%, respectively, with a 50% reduction in meat and dairy consumption in Europe (Westhoek et al., 2014).

Furthermore, the steady global population growth and industrial development have resulted in an increased demand for food and meat production (Flynn et al., 2019). In the context of achieving global food security, the increasing rate of meat consumption expected for the following years will be contentious to maintain (Hicks et al., 2018). This arises an urgent need for new protein-rich alternatives to animal food in human nutrition (Fasolin et al., 2019). The main problem is that animal protein production is poorly efficient compared to plant protein (Nadathur et al., 2017), which can be estimated by comparing the grams of protein delivered to the wholesale point per unit energy used or per unit total greenhouse gases. About 30% of the global land surface (7% for feed and 23% of pasture) is needed for animal production to meet humans' food needs, while about 7% is used for cropland for human consumption (Stoll-Kleemann and O'Riordan, 2015; Alexander et al., 2016; Salter, 2018).

The health, environmental, and food security problems associated with an excess of meat production and consumption have seemed to be appealing to a segment of Western meat-eaters to adopt specific meat consumption approaches aimed at reducing the amount of meat in their diets (Hicks et al., 2018; Sánchez-Sabaté and Sabaté, 2019). Among them, an emerging approach is to diversify the food types in the diet in order to contribute to more sustainable use of meat and consumption habits, while achieving a high nutritional value. This means a trend toward flexitarian diets (Derbyshire, 2017; Hicks et al., 2018).

Considering the above-mentioned concerns and consumers' tendencies and regarding the processed meat industry, reformulation strategies are needed in order to obtain either healthier processed meat or reduced-meat products. Approaches to deliver healthier meat products have been aimed at fat, sodium, or nitrite reduction, fatty acid profile modification, and inclusion of calcium, vitamins, and nutritional, functional ingredients such as fiber or phytochemicals (Grasso et al., 2014; Thøgersen and Bertram, 2021). Strategies to deliver reduced meat products have been based on a partial replacement of meat (muscle protein) with vegetal ingredients with high protein content, with the suggested ingredients being grain legumes, cereals, oilseeds, or mushrooms or protein concentrates obtained from them (Asgar et al., 2017).

REFORMULATION APPROACHES FOR REDUCED-MEAT HEAT-TREATED EMULSION-TYPE SAUSAGES

Muscle proteins exert different technological–functional properties, which are crucial for the edible quality of the resulting meat products, i.e., viscoelastic textural characteristics responsible for their chewiness and mouthfeel (Sha and Xiong, 2020). The muscle proteins with higher functionality in meat products are myofibrillar proteins, mainly myosin, and collagen (Asgar et al., 2017). In the making process, thanks to the ionic strength provided by the common salt and other salts such as phosphates and the cutter's mechanical energy, a large amount of the myofibrillar proteins are solubilized. The proteins activated immobilize water, interact among them, and with the hydrocolloids, e.g., starch or carrageenan, which are eventually used as ingredients, and cover and emulsify the small fat particles formed. Moreover, when the sausage is cooked, proteins, and other gelling agents form a high viscoelastic gel (Sikorski, 2001; Sha and Xiong, 2020).

The partial replacement of the muscle proteins by non-meat proteins, such as soy protein, caseinates or gluten, pork skin or gelatin, starches or gums, has been standard in the emulsion-type meat sausage-making process (Santhi et al., 2015). This practice's primary purpose is to reduce the cost of raw materials, being usually the meat the more expensive raw material (Feiner, 2006). According to this author, sausages with non-muscle protein and meat extenders can show an acceptable cost/eating quality ratio; however, if a large amount of meat is replaced, the nutritive value of sausage can be significantly reduced.

The reformulation of emulsion-type sausages toward meat reduction cannot only be directed to reduce the costs of raw materials, but it can also be primary aimed at making sausages healthier. The main approaches used to accomplish this goal include the use of vegetable oils as animal fat replacers or the utilization of phytochemical- or dietary fiber-rich ingredients in the formulations (Weiss et al., 2010; Kaur and Sharma, 2019; Das et al., 2020). Moreover, reformulation can also reduce the meat content and consequently diminish the environmental effect associated with excessive meat production. In this approach, the critical question is to obtain sausages with lower meat content (reduced-meat products), as nutritious as the conventional meat products or even more, and showing sensory characteristics quite similar to those of the conventional meat products in order to avoid rejection by frequent meat consumers (Hoek et al., 2011).

TABLE 1 | Amounts (g) of food or food protein to provide the recommended daily allowance for the corresponding limiting essential amino acid, referred to a 70-kg man, considering a protein content of 21, 11, 12, 11, 7, and 2% for pork, egg, beans, wheat, rice, and potato, respectively.

	Pork	Egg	Beans	Wheat	Rice	Potato
Food	137	206	267	339	439	2,063
Food protein	28	25	32	37	31	41
Limiting amino acid	–	–	Meth/Cys	Lys	Lys	iso-Leu

Adapted from Tessari et al. (2016).

PULSES AS CANDIDATE INGREDIENTS TO PREPARE REDUCED-MEAT PRODUCTS

Dry grain legumes or pulses, such as lentils, chickpeas, or beans, are foods produced and consumed in large amounts worldwide and are considered relatively cheap healthy food in most regions (Headey and Alderman, 2019). Furthermore, they can be used for intercropping and cover crops, thus enhancing their production sustainability (Maitra et al., 2021). As for their nutritional value, pulses contain high levels of proteins (20–30%), minerals, B-group vitamins, and dietary fiber, apart from several phytochemicals (Farooq and Boye, 2011). *In vitro* protein digestibility of pulse flours has been reported to be around 80% (Bessada et al., 2019) and their essential amino acid index ranged 50–65% (Khattab et al., 2009). The protein quality of pulses as regards to the essential amino acid content is relatively high as compared with cereals, however, it is worse than that of meat (**Table 1**), which is due to a low content in sulfur amino acids and tryptophan, the limiting amino acids in pulse protein (Iqbal et al., 2006; Boye et al., 2010; Grela et al., 2017).

Nonetheless, when pulses are combined with rice or egg protein, the protein biological value considerably improves (Farooq and Boye, 2011). Pulses also contain a large amount of starch (50–70%), composed of amylose and amylopectin chains. Its high resistance to digestion confers the pulse starch a dietary interest since it acts as dietary fiber (Singh et al., 2017).

Pulses, or proteins extracted from them, have been suggested as ingredients to be considered in reformulation of processed meat. The principal proteins in pulses are albumins and globulins. The firsts are water-soluble and are composed of enzymes, lectins, enzymatic inhibitors. Globulins, also known as reserve proteins, are the most abundant and are soluble in saline solutions (Singhal et al., 2016). These proteins are the main ones responsible for functionality, i.e., water retention, emulsion, gel formation, attributed to pulse proteins (Farooq and Boye, 2011; Toews and Wang, 2013; Shevkani et al., 2017; Jarpa-Parra, 2018). Their functional performance is comparable or even higher to soy protein, which is frequently used in the meat industry.

The pulses' starch forms gels at temperatures between 70 and 90°C (Farooq and Boye, 2011). When pulses are mixed with other ingredients, for making bakery, pasta, food emulsions, or meat products, both proteins and starches establish chemical links and form structures of different degree of firmness depending on the food matrix pH, ionic strength, gelling agent concentrations, fat content, or heating conditions (Sozer et al., 2017).

A relevant problem derived from the utilization of pulse flours as ingredients in processed food is that pulses contain a high amount of natural antinutrient compounds necessary to reduce (Paterson et al., 2017). These can be classified according to their composition in non-proteinaceous and proteinaceous. Among the first type are phytic acid, oxalate, saponins, and cyanogenic glycosides that can interact with food proteins, mineral, vitamins, or carbohydrates. The second group is composed of lectins, agglutinins, trypsin, chymotrypsin, and amylase inhibitors that interact with human enzymes or can agglutinate red blood cells (Sreerama et al., 2012).

Different pulse processing techniques, such as dehulling, soaking, extrusion, micronization, heating, germination, or fermentation, have been found to reduce the content of antinutrient compounds in pulse flours (Kumar et al., 2009; Paterson et al., 2017; Kaspchak et al., 2018). Dehulling mainly decreases the tannin content in seeds, between 68 and 90%, and improves their nutritional quality (Kumar et al., 2009). Soaking is effective in reducing the tannin and phytic acid contents to an extent depending on soaking conditions, i.e., time and temperature combinations, and pulse type (Martín-Cabrejas et al., 2009; Paterson et al., 2017). The reduction tends to be higher when pulses are soaked in salt solutions (Taiwo and Akanbi, 1997). Heat treatment considerably reduces phytic acid and tannin contents and is the most effective process to reduce trypsin inhibitors (Osman, 2007; López-Martínez et al., 2017); however, due to their remarkable heat stability, an intense heating treatment is needed, e.g., boiling or autoclaving, to reduce most of the activity (Chan et al., 2014). Finally, germination and fermentation improve the digestibility and the availability of certain nutrients, i.e., minerals, and reduce the levels of antinutrient compounds, such as phytic acid, tripsin inhibitors, and α-galactosides; however, the reduction of enzyme inhibitors by these processes is lower than boiling or autoclaving (Kumar et al., 2009; Wood and Malcolmson, 2011; Gharachorloo et al., 2013; Paterson et al., 2017).

RESEARCH ON THE UTILIZATION OF PLUSES AS MEAT REPLACERS IN PROCESSED MEAT

The research on the utilization of pulse flour as a meat replacer is not new. As a matter of example, Verna et al. (1984) studied the effect of the partial replacement of meat with chickpea flour, based on a protein-to-protein replacement, on the microbial quality of English-type fresh sausages prepared from either mutton, pork, or beef. The technological interest in the use of pulses in the meat industry keeps active. Recent research studies on the reformulation of minced-meat preparations such as burgers, patties, meatballs, and fresh sausages with pulses as meat replacers can be found in the literature (Serdaroglu et al., 2005; Holliday et al., 2011; Ghribi et al., 2018; Argel et al., 2020; Pintado and Delgado-Pando, 2020). In these studies, pulse flour was used at levels between 5 and 50%, with a reduction in the meat content from c.a. 5 to 40%. Results have shown, in general, that the pulse flour at amounts lower than 15% increased the cooking yield and firmness of the meat preparations without exerting, in most cases, a negative effect on the sensory acceptation. Furthermore, it has been found that the effect of flour pulses on the quality of reduced meat preparations not only depends on the flour amount used, and the amount of meat that was replaced, but also on the type of pulse (Holliday et al., 2011).

However, research on the use of pulses in emulsion-type cooked meat products seems to be scarce. Dzudie et al. (2002), Sanjeewaa et al. (2010), and Albarracín et al. (2010) prepared emulsion-type sausages with bean or chick pea flours at levels up to 10%, and a subsequent meat reduction equal to pulse flour added. Furthermore, Tahmasebi et al. (2016) formulated sausages with variable amounts of pigeon pea flour (up to 22%), combined with corn flour, walnut, and sesame paste. A summary of the methodology and results from those studies found are shown in **Table 2**. In general, the use of pulse flour increased sausage yield and emulsion stability and increased lightness (L^*). Regarding texture, the four studies reported increased cohesiveness due to pulse flour and three out of the four higher hardness, although an inverse effect was observed in the other study (Dzudie et al., 2002). Finally, the effect of pulse flour on consumers' acceptance was not consistent, i.e., while Albarracín et al. (2010) reported lower acceptance scores for Frankfurters with pulse flour, (Sanjeewaa et al., 2010) found no negative effect of pulse flour on the sausage flavor.

REFORMULATION CHALLENGES TO INCLUDE PULSES IN HEAT-TREATED EMULSION-TYPE SAUSAGES

Due to the scarcity of scientific information, further research on the reformulation of emulsion-type meat sausages using pulses is deserved, and the lines to explore are many. As relevant research, the effects of different levels of meat (muscle proteins) replacement by pulse flour (pulse proteins) on the sausage quality traits should be studied. In these reformulations, the amounts of protein, water, and starch should be optimized. Furthermore, the incorporation of pulses could be combined with other functional proteins such as collagen or albumin, which could improve the technological functionality and the sausage edible quality and nutritive value. The response surface methodology and mixture design could be used as a valuable tool to select the best formulations (Arteaga et al., 1994). Moreover, the flour of different pulses should be tested, and their performance compared among them.

Previous to the research mentioned above, suitable procedures to prepare the pulse flours and incorporate them in the sausage-making process (pulse flour pre-treatment) should be researched. Pulse flour functional properties such as protein solubility, water absorption, or pasting properties, among others, would depend on pulse pre-treatments such as soaking, dehulling, germination, or heating (Felker et al., 2003; Wood and Malcolmson, 2011; Ribéreau et al., 2018). Furthermore, functional properties of pulse flours as food ingredients are significantly affected by flour particle size; thus the particle size should be appropriate for the product concerned (Wood and Malcolmson, 2011).

Moreover, before adding the pulse flour to the sausage batter, mixing or homogenizing pulse flour in water or saline could improve pulse protein and starch functionality. This statement's rationale regarding proteins is that globulins, the more significant proteins in pulses, are highly soluble in saltwater (Bessada et al., 2019). It might be advantageous to solubilise these proteins before incorporating the pulse flour into the sausage batter.

Regarding pulse antinutrients, it should be relevant to study to what extent pulse or pulse flour pre-treatments, i.e., soaking, germination, toasting, or the heat-treatment intensity of the pulse-containing sausages, would inactivate or remove those compounds. On the other hand, it should be investigated how the pre-treatment, supposing pulses are pre-treated to diminish their antinutrient content (Bessada et al., 2019), could affect pulse proteins' technological functionality and how it could affect the edible sausage yield and texture quality. Furthermore, the increment in the cost of using processed legumes instead of non-treated pulses should also be considered.

The effect of relevant sausage processing conditions such as the amount of phosphates required, the optimum cuterization time or the heating intensity warrant further research. For the latter, it should be considered that gelatinization temperature of pulse starch is higher (70–90°C; Farooq and Boye, 2011) than that of the conventionally used potato starch (about 60°C; Feiner, 2006).

Possible adverse effects of pulse flour on the quality of emulsion-type sausages might be manifested, and, in this case, the defects should be addressed with appropriate approaches. The high content of fiber in pulses might result in a less accepted texture, e.g., excessive hardness or atypical mouthfeel, compared to conventional sausages. In this respect, the use of flours from dehulled pulses instead of wholemeal flour, can be considered to reduce fiber content (Wood and Malcolmson, 2011). Pulse flours could also be accompanied with collagen to improve mouthfeel. In this regard, Hjelm et al. (2019) found the use of collagen protein (1–3 mg/100 g) to ameliorate the negative effect of the inclusion of fiber (rye bran) on Frankfurter texture. Nonetheless, fibre's adverse effect might not be detected since Cofrades et al. (2000) did not found fiber (2% oat fiber) to affect the texture of emulsion-type sausages.

Furthermore, the effect of pulses in the sausage color might diminish its color acceptance, and it could also exert an eventual negative effect on flavor. Pulses could impart the sausage an undesirable "beany" flavor (Bessada et al., 2019) or promote lipid oxidation due to the action of lipoxidase, which seems to be able to resist the sausage pasteurization heating treatment (Wood and Malcolmson, 2011). On the other hand, pulses such as lentils are rich in polyphenols, thus being a source of natural antioxidants (Munekata et al., 2020), thus improving the oxidative stability of sausages. Besides, the effects of incorporating pulse flour on microbial concentration, microbial growth, and sausage shelf life also deserve to be studied and understood.

In conclusion, the utilization of pulse flours in the formulation of reduced-meat heat-treated emulsion meat sausages without resulting in an adverse effect on the sausage quality seems

TABLE 2 | Methodology and results from studies testing the effect pulse flours on the quality of emulsion-type sausages.

Study	Albarracín et al., 2010	Dzudie et al., 2002	Sanjeewaa et al., 2010	Tahmasebi et al., 2016
Pulse type	Beans (*Phaseolus* spp.)	Common bean (*Phaseolus vulgaris*)	Chickpea (*Cicer arietinum*)	Pigeon pea (*Cajanus cajan*)
Pre-treatment	Soaked, cooked (155°C, 30 min), mashed and dried	Dehulled	No pretreatment	No-pretreatment
Particle size	0.08 mm	40 mesh∼0.2 mm	0.1 mm	0.75 mm
Sausage type	Frankfurter	Low-fat emulsion-type sausage	Low-fat bologna	Low-fat emulsion-type sausage
Pulse flour amount	0–9%	0–10%	0–5%	0–22%
Meat replacement	Meat replaced with pulse flour at a ratio 1:1	Meat replaced with pulse flour at a ratio 1:1	Meat replaced with pulse flour at a ratio 1:1	Meat and fat replaced with pulse flour combined with corn flour, walnut, and sesame paste[a]
Heating (core temperature)	72°C	72°C	72°C	80°C
Effect on color	Decreased a* and increased b* and L*	Increased L* at levels of 5.0 and 10.0%.		
Effect on yield	–	Increased water retention	Increased water retention	Increased water retention and emulsion stability
Effect on texture	At the highest level increased hardness	Decreased hardness and increased cohesiveness.	Increased hardness/firmness	Increased hardness
Effect on sensory quality	Consumer acceptance decreased as pulse flour level increased	–	Pulse flour up to 5% was not detected by the panelists (flavor)	–

[a] *Formulated using response surface methodology.*

feasible. Nonetheless, further research is warranted to develop best practices and strategies. This research should aim to prevent the presence of dangerous levels of pulse antinutrients, achieve a high pulse protein functionality and search for suitable combinations of pulse flour with proteins, i.e., collagen or albumin, or spices and condiments to improve the quality of pulse-containing sausages.

AUTHOR CONTRIBUTIONS

JM: conception, compiling references, writing, drafting, and coordination. IC, SK, and AC: compiling references and writing. BS: conception, compiling references, and writing. DR: funding, conception, writing, and coordination. All authors contributed to the article and approved the submitted version.

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